

Figure 1 (SEQ ID NO:724)

85P1B3 SSH sequence and GenBank homology to OIP5

GATCAGAGGACACATGGGACTCTGCATCTTAATTCCTAAATTTACAGTCAAAGACATTTTCAG
 AGATAAGTATTATGAATTCAATAAGAATCTAAAGTAAGTTCTTAAGGCAAATAGCTATAAAA
 GAGAAGAATCCTTAGTCTCTCATCTTCTAAAAACAGCTTCACAAATAATTTGGAAAATCAGCC
 TAAAGGTAAATAGAACTGCATTTCCCCTCCATTCTTGAAGCCAATCTTTTCAAGAAATGAC
 TAAGCAGCACCTGTTGTTGAAGACAGCAATAAAGCCTGAACCTGACACTCAAGCTTTGGTACA
 GGATC

gb|AF025441.1|AF025441 Homo sapiens Opa-interacting protein... 632 e-179
 gb|AF158642.1|AF158642 Homo sapiens metalloproteinase-disin... 42 0.12
 gb|AC005075.2|AC005075 Homo sapiens clone RG219E16, complet... 42 0.12
 emb|AL096773.6|HS1000E10 Human DNA sequence from clone 1000... 40 0.48

>gb|AF025441.1|AF025441 Homo sapiens Opa-interacting protein OIP5 mRNA, partial cds
 Length = 1197

Score = 632 bits (319), Expect = e-179
 Identities = 319/319 (100%)
 Strand = Plus / Minus

Query: 1 gatcagaggacacatgggactctgcatcttaattcctaaatttacagtcaaagacatttt 60
 ||||||||||||||||||||||||||||||||||||||||||||||||||||||||
 Sbjct: 1013 gatcagaggacacatgggactctgcatcttaattcctaaatttacagtcaaagacatttt 954

Query: 61 cagagataagtattatgaattcaataagaatctaaagtaagttcttaaggcaaataagcta 120
 ||||||||||||||||||||||||||||||||||||||||||||||||||||||||
 Sbjct: 953 cagagataagtattatgaattcaataagaatctaaagtaagttcttaaggcaaataagcta 894

Query: 121 taaaagagaagaatccttagtctctcatcttctaaaaacagcttcacaaataatttgga 180
 ||||||||||||||||||||||||||||||||||||||||||||||||||||||||
 Sbjct: 893 taaaagagaagaatccttagtctctcatcttctaaaaacagcttcacaaataatttgga 834

Query: 181 aatcagcctaaaggtaaataagaactgcatttcccctccattcttgaagccaatcttttt 240
 ||||||||||||||||||||||||||||||||||||||||||||||||||||||||
 Sbjct: 833 aatcagcctaaaggtaaataagaactgcatttcccctccattcttgaagccaatcttttt 774

Query: 241 caagaaatgactaagcagcacctgttggtgaagacagcaataaagcctgaacctgacact 300
 ||||||||||||||||||||||||||||||||||||||||||||||||||||||||
 Sbjct: 773 caagaaatgactaagcagcacctgttggtgaagacagcaataaagcctgaacctgacact 714

Query: 301 caagctttggtacaggatc 319 (SEQ ID NO:725)
 ||||||||||||||||
 Sbjct: 713 caagctttggtacaggatc 695 (SEQ ID NO:726)

Figure 2

cDNA Sequence and ORF of 85P1B3/OIP5 clone A
(SEQ ID NO:727)

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5' 9      18      27      36      45      54
   GGC TGC GGG AAG ATG GCG GCT CAG CCG CTG CGG CAT CGC TCA CGT TGT GCA ACG
   -----
   (SEQ ID NO:728)→ M  A  A  Q  P  L  R  H  R  S  R  C  A  T

   63      72      81      90      99      108
   CCG CCC CGG GGG GAC TTT TGT GGT GGC ACT GAG AGG GCG ATT GAC CAA GCT TCT
   -----
   P  P  R  G  D  F  C  G  G  T  E  R  A  I  D  Q  A  S

   117     126     135     144     153     162
   TTT ACG ACC TCC ATG GAG TGG GAT ACG CAG GTG GTG AAG GGG TCC TCG CCG CTC
   -----
   F  T  T  S  M  E  W  D  T  Q  V  V  K  G  S  S  P  L

   171     180     189     198     207     216
   GGC CCC GCA GGG CTG GGG GCT GAG GAG CCA GCC GCC GGC CCG CAG CTG CCG TCT
   -----
   G  P  A  G  L  G  A  E  E  P  A  A  G  P  Q  L  P  S

   225     234     243     252     261     270
   TGG CTG CAG CCT GAG AGG TGC GCT GTG TTC CAG TGC GCA CAG TGT CAC GCA GTG
   -----
   W  L  Q  P  E  R  C  A  V  F  Q  C  A  Q  C  H  A  V

   279     288     297     306     315     324
   CTC GCC GAC TCG GTG CAC CTC GCC TGG GAC CTG TCG CGG TCC CTC GGG GCC GTG
   -----
   L  A  D  S  V  H  L  A  W  D  L  S  R  S  L  G  A  V

   333     342     351     360     369     378
   GTC TTC TCC AGA GTT ACA AAT AAC GTC GTT TTG GAA GCG CCC TTC CTA GTT GGC
   -----
   V  F  S  R  V  T  N  N  V  V  L  E  A  P  F  L  V  G

   387     396     405     414     423     432
   ATT GAA GGT TCA CTC AAA GGC AGT ACT TAC AAC CTT TTA TTC TGT GGT TCT TGT
   -----
   I  E  G  S  L  K  G  S  T  Y  N  L  L  F  C  G  S  C

   441     450     459     468     477     486
   GGG ATT CCC GTT GGT TTC CAT CTG TAT TCT ACC CAT GCT GCC CTG GCT GCC TTG
   -----
   G  I  P  V  G  F  H  L  Y  S  T  H  A  A  L  A  A  L

   495     504     513     522     531     540
   AGA GGT CAC TTC TGC CTT TCC AGT GAC AAA ATG GTG TGC TAT CTC TTA AAA ACA
   -----
   R  G  H  F  C  L  S  S  D  K  M  V  C  Y  L  L  K  T

   549     558     567     576     585     594
   AAA GCC ATA GTA AAT GCA TCA GAG ATG GAT ATT CAA AAT GTT CCT CTA TCA GAA
   -----
   K  A  I  V  N  A  S  E  M  D  I  Q  N  V  P  L  S  E

   603     612     621     630     639     648
   AAG ATT GCA GAG CTG AAA GAG AAG ATA GTG CTA ACG CAC AAT CGC TTA AAA TCA
   -----
   K  I  A  E  L  K  E  K  I  V  L  T  H  N  R  L  K  S

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702

657				666				675				684				693				702			
CTA	ATG	AAG	ATT	CTG	AGT	GAA	GTG	ACT	CCT	GAC	CAG	TCC	AAG	CCA	GAA	AAC	TGA						
L	M	K	I	L	S	E	V	T	P	D	Q	S	K	P	E	N	*						
711				720				729				738				747				756			
TCC	TGT	ACC	AAA	GCT	TGA	GTG	TCA	GGT	TCA	GGC	TTT	ATT	GCT	GTC	TTC	AAC	AAC						
765				774				783				792				801				810			
AGG	TGC	TGC	TTA	GTC	ATT	TCT	TGA	AAA	AGA	TTG	GCT	TCA	AGA	ATG	GAG	GGG	AAA						
819				828				837				846				855				864			
TGC	AGT	TTC	TAT	TTA	CCT	TTA	GGC	TGA	TTT	TCC	AAA	TTA	TTT	GTG	AAG	CTG	TTT						
873				882				891				900				909				918			
TTA	GAA	GAT	GAG	AGA	CTA	AGG	ATT	CTT	CTC	TTT	TAT	AGC	TAT	TTG	CCT	TAA	GAA						
927				936				945				954				963				972			
CTT	ACT	TTA	GAT	TCT	TAT	TGA	ATT	CAT	AAT	ACT	TAT	CTC	TGA	AAA	TGT	CTT	TGA						
981				990				999				1008				1017				1026			
CTG	TAA	ATT	TAG	GAA	TTA	AGA	TGC	AGA	GTC	CCA	TGT	GTC	CTC	TGA	TCT	AAA	GTT						
1035				1044				1053				1062				1071				1080			
GCA	TGG	TTG	GTC	TGA	AAA	TAG	AGT	TGG	GCT	TAA	TGT	TGA	CTT	CTA	TTA	CTC	CTG						
1089				1098				1107				1116				1125				1134			
CAT	GGA	GCA	GTT	GTT	ATG	AAT	ACT	AAT	ACA	TCA	CTT	TTT	AAC	TTC	TGT	AAA	ATA						
1143				1152				1161				1170				1179				1188			
CAG	ATC	ATA	ATA	TTC	TAT	AGG	TAA	TGT	TTA	ATA	AAT	TGC	CTG	AAT	AAT	AAA	AAA						
1197				1206				1215				1224				1233				1242			
AAA	AAA	AAA	AAA	AAA	AAA	AAA	AAA	AAA	AAA	AAA	AAA	AAA	AAA	AAA	AAA	AAA	AAA						
1251				1260																			
AAA	AAA	AAA	AAA	AAA	AAA	AA	3'																

Figure 3

85P1B3/OIP5 protein sequence. (SEQ ID NO:729)

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1  MAAQPLRHRS RCATPPRGDF CGGTERAIDQ ASFTTSMEWD TQVVKGSSPL GPAGLGAEEL
61  AAGPOLPSWL QPERCAVFQC AQCHAVLADS VHLDLWLSRS LGAVVFSRVT NNVLEAPFL
121 VGIEGSLKGS TYNLLFCGSC GIPVGFHLYS THAALAALRG HFCLSSDKMV CYLLKTKAIV
181 NASEMDIQNV PLSEKIAELK EKIVLTHNRL KSLMKILSEV TPDQSKPEN*

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Figure 4

Alignment of 85P1B3 with OIP5.

>gi|2815610|gb|AAC39561.1| (AF025441) Opa-interacting protein OIP5 [Homo sapiens]
Length = 231

Score = 462 bits (1189), Expect = e-130

Identities = 229/229 (100%), Positives = 229/229 (100%)

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85P1B3: 1  MAAQPLRHRSRCATPPRGDFCGGTERAIDQASFTTSMEDWTQVVKGSSPLGPAGLGAEED 60
      MAAQPLRHRSRCATPPRGDFCGGTERAIDQASFTTSMEDWTQVVKGSSPLGPAGLGAEED
OIP5: 3  MAAQPLRHRSRCATPPRGDFCGGTERAIDQASFTTSMEDWTQVVKGSSPLGPAGLGAEED 62

85P1B3: 61  AAGPQLPSWLQPERCAVFQCAQCHAVLADSVHLAWDLRSISLGAVVFSRVTTNNVVLEAPFL 120
      AAGPQLPSWLQPERCAVFQCAQCHAVLADSVHLAWDLRSISLGAVVFSRVTTNNVVLEAPFL
OIP5: 63  AAGPQLPSWLQPERCAVFQCAQCHAVLADSVHLAWDLRSISLGAVVFSRVTTNNVVLEAPFL 122

85P1B3:121  VGIEGSLKGSTYNLLFCGSCGIPVGFHLYSTHAALAALRGHFCLSSDKMVCYLLKTKAIV 180
      VGIEGSLKGSTYNLLFCGSCGIPVGFHLYSTHAALAALRGHFCLSSDKMVCYLLKTKAIV
OIP5: 123  VGIEGSLKGSTYNLLFCGSCGIPVGFHLYSTHAALAALRGHFCLSSDKMVCYLLKTKAIV 182

85P1B3:181  NASEMDIQNVPLSEKIAELKEKIVLTHNRLKSLMKILSEVTPDQSKPEN 229 (SEQ ID NO:730)
      NASEMDIQNVPLSEKIAELKEKIVLTHNRLKSLMKILSEVTPDQSKPEN
OIP5: 183  NASEMDIQNVPLSEKIAELKEKIVLTHNRLKSLMKILSEVTPDQSKPEN 231 (SEQ ID NO:731)

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Figure 5: 85P1B3 Hydrophilicity profile
(Hopp T.P., Woods K.R., 1981. Proc. Natl. Acad. Sci. U.S.A. 78:3824-3828)

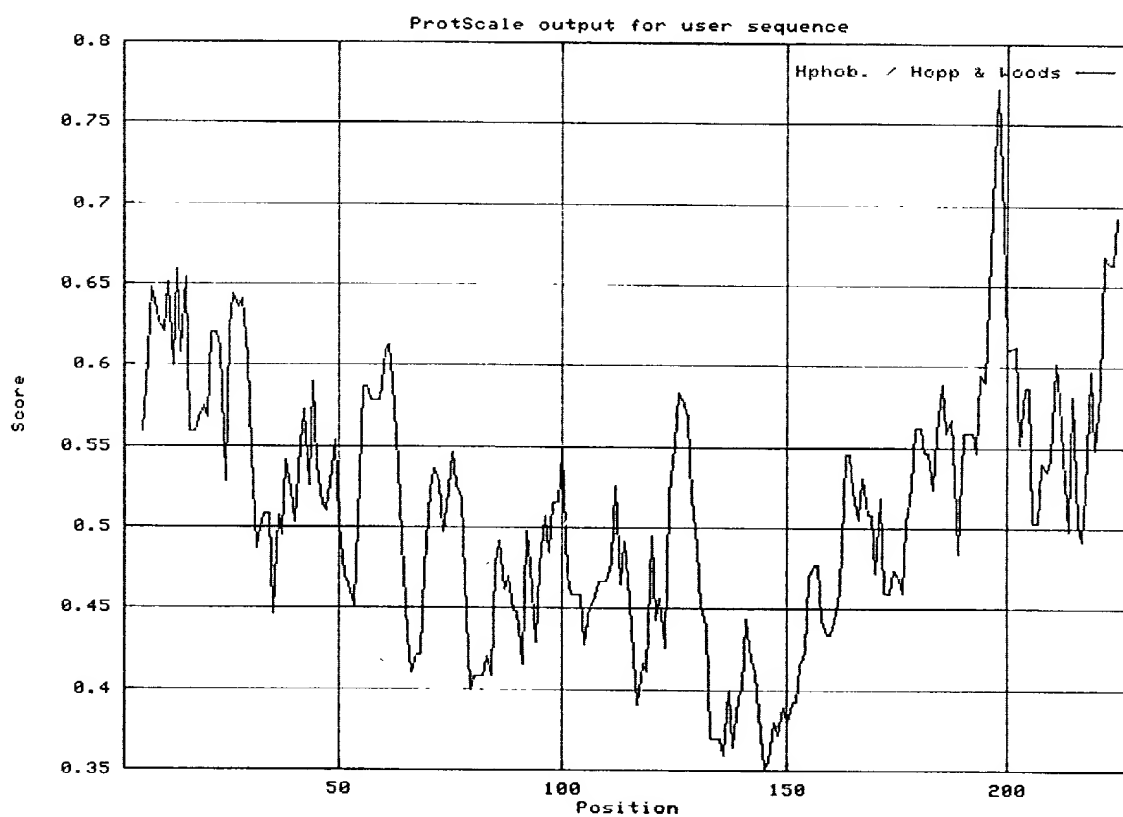


Figure 6: 85P1B3 Hydropathicity Profile
(Kyte J., Doolittle R.F., 1982. J. Mol. Biol. 157:105-132)

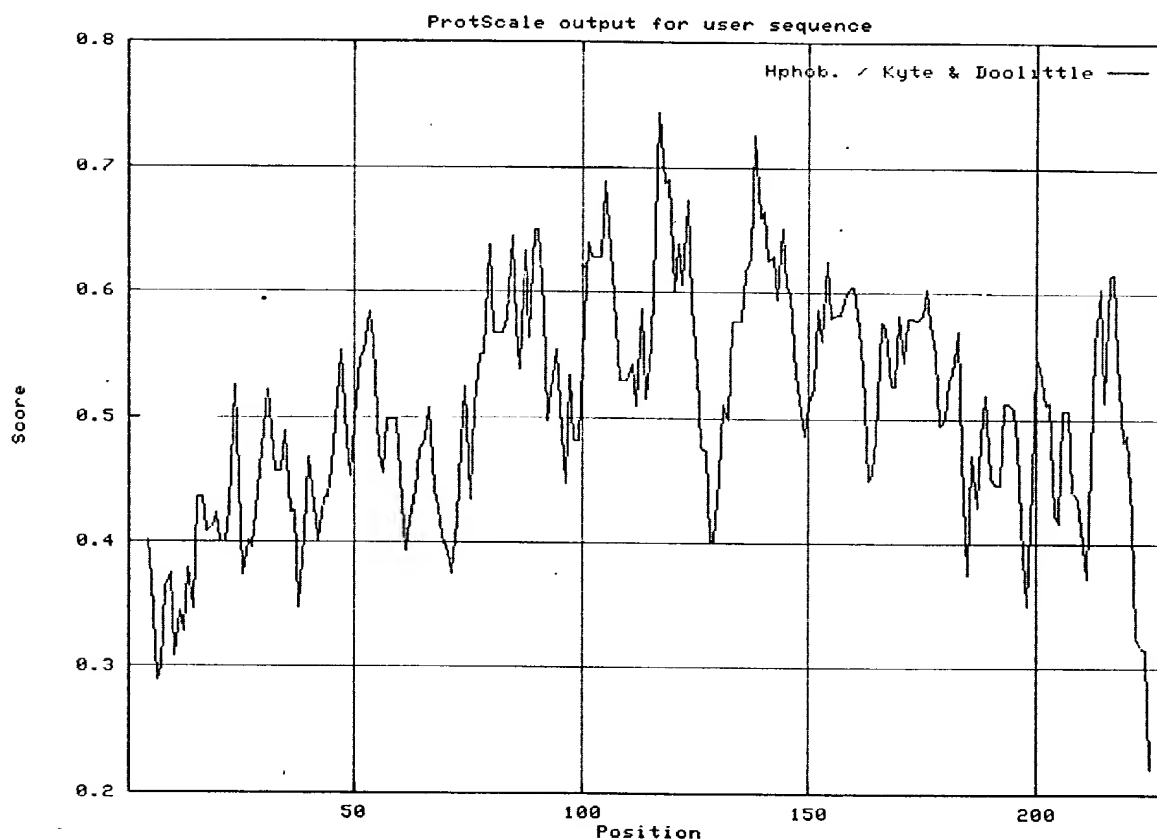


Figure 7: 85P1B3 % Accessible Residues Profile
(Janin J., 1979. Nature 277:491-492)

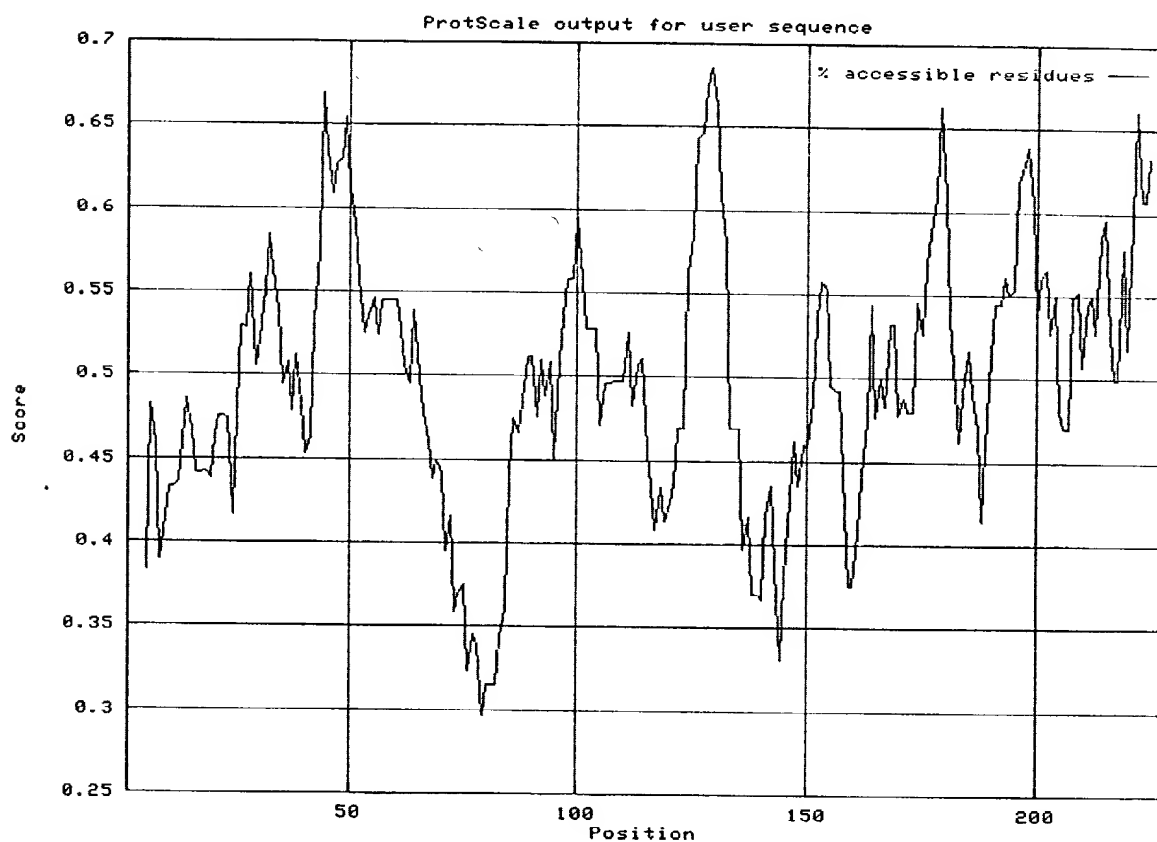


Figure 8: 85P1B3 Average Flexibility Profile

(Bhaskaran R., Ponnuswamy P.K., 1988.

Int. J. Pept. Protein Res. 32:242-255)

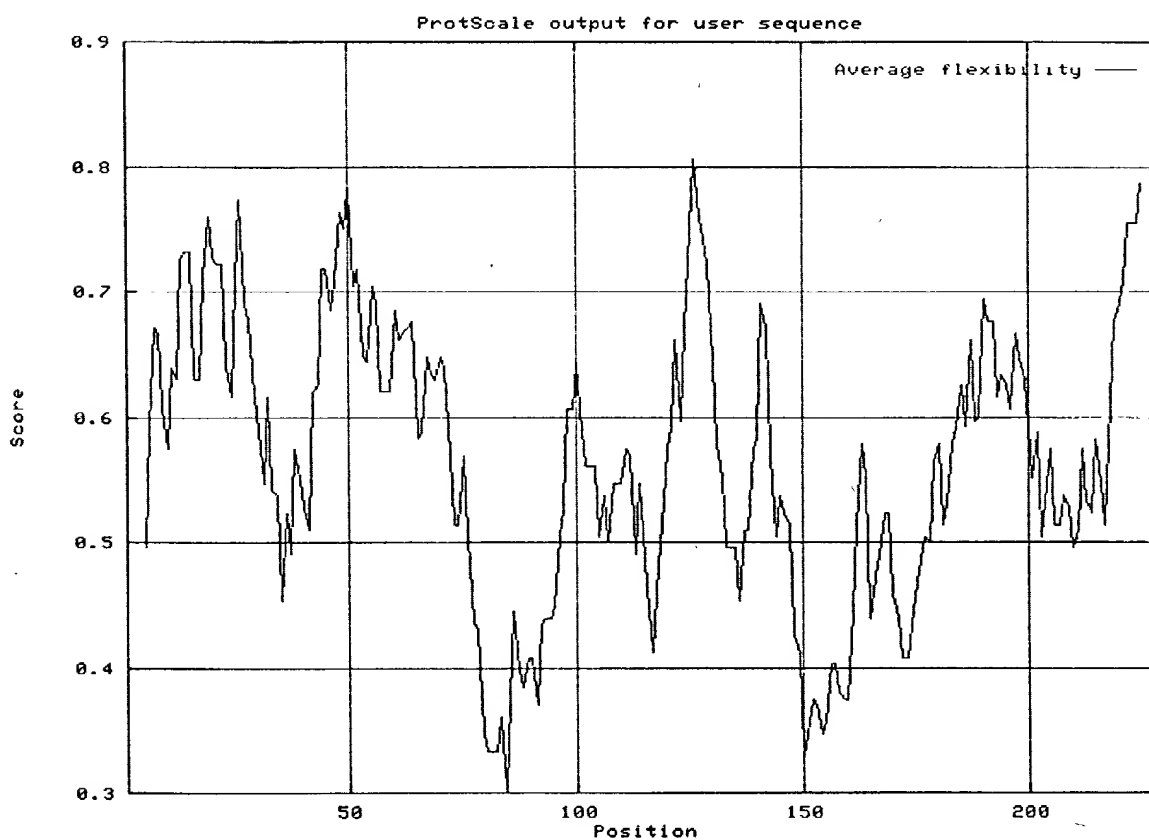


Figure 9: 85P1B3 Beta-turn Profile
(Deleage, G., Roux B. 1987. Protein Engineering 1:289-294)

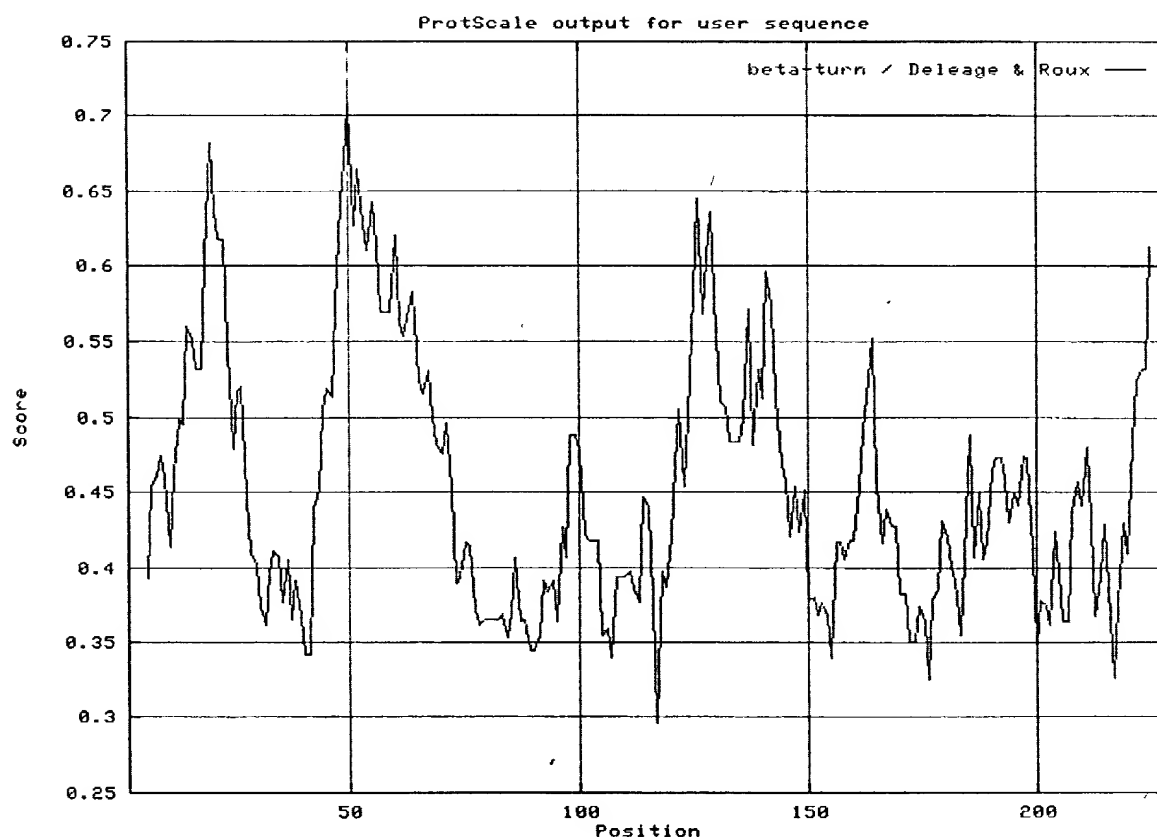


Figure 10 RT-PCR analysis of 85P1B3 expression.

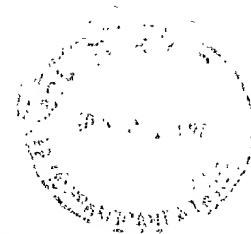
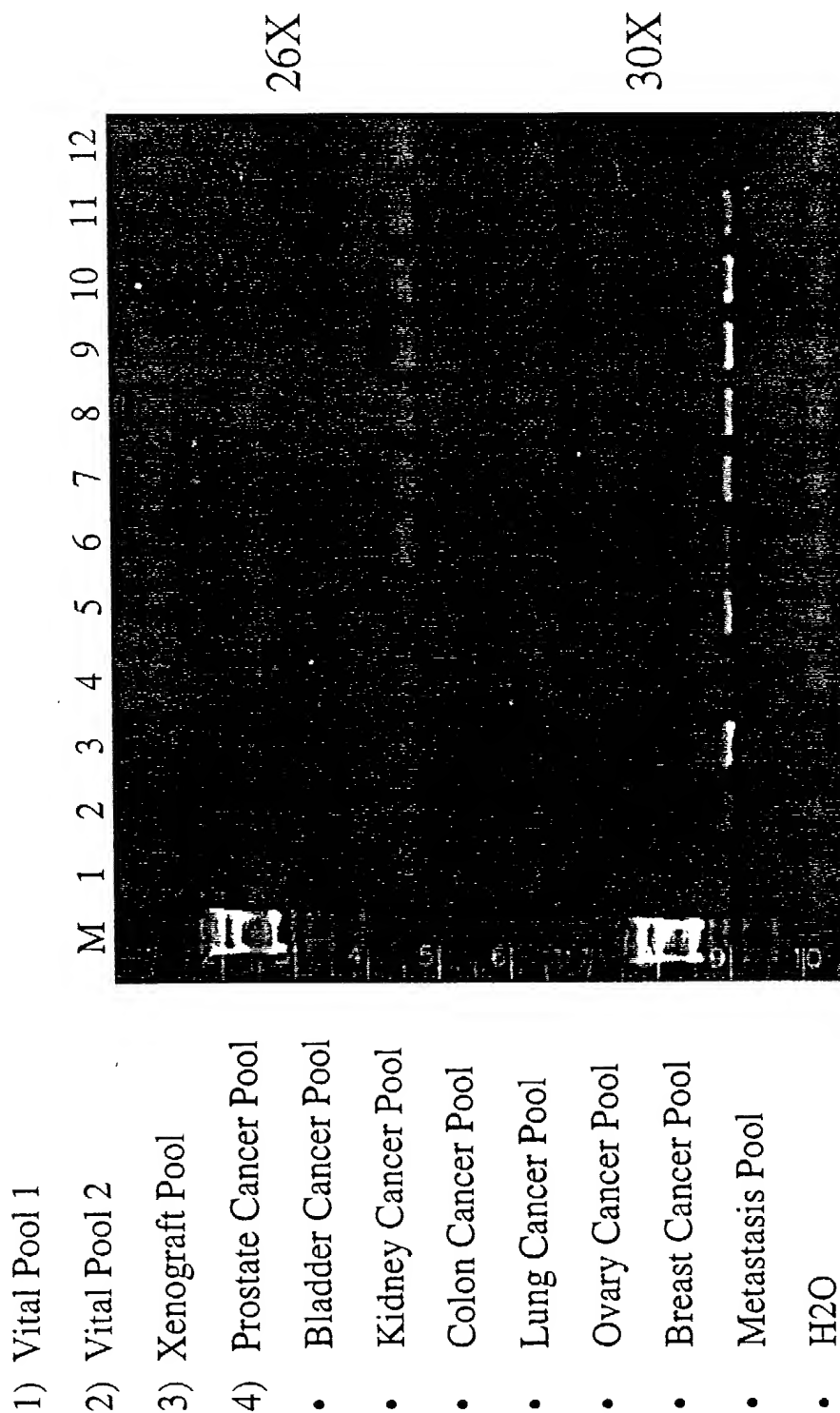


Figure 11 Expression of 85P1B3 in Normal Human Tissues

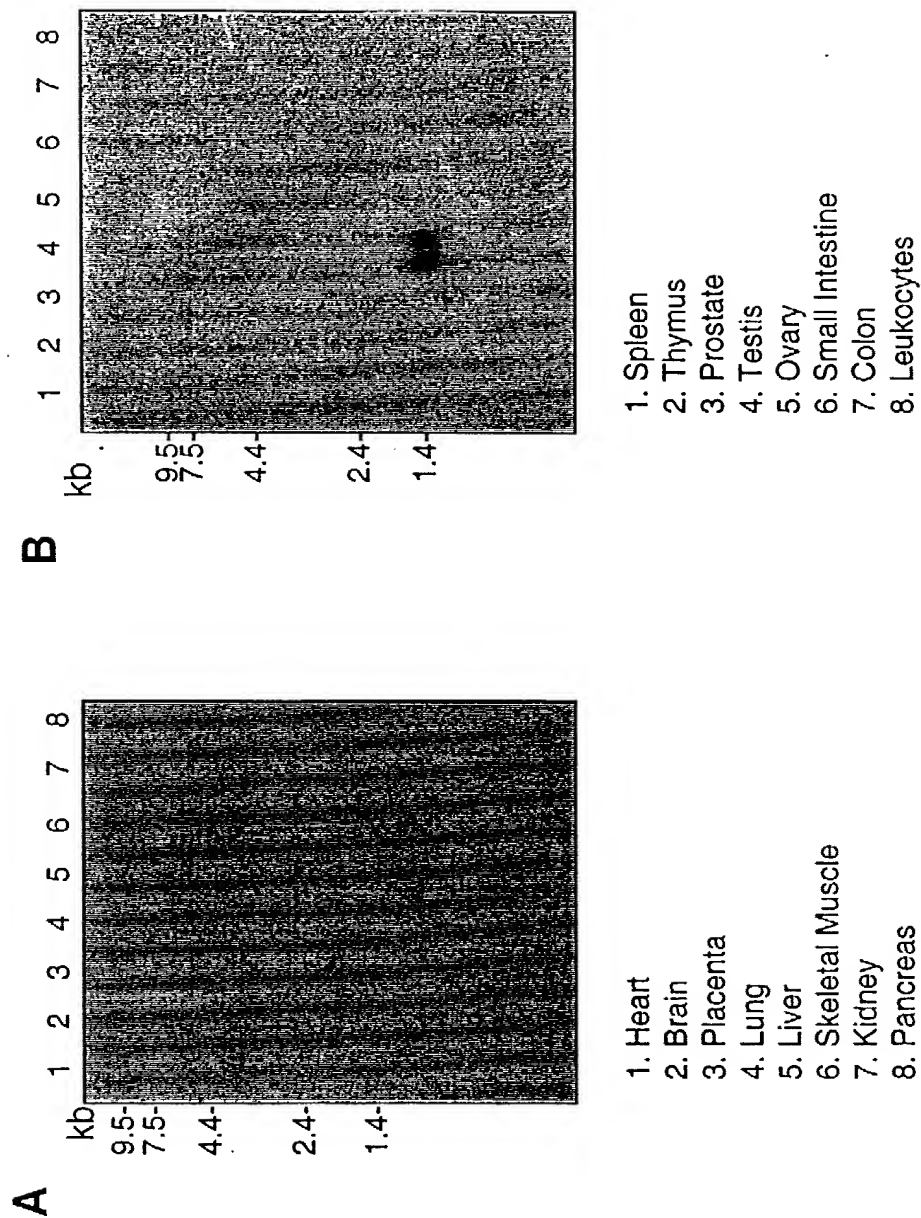


Figure 12 Expression of 85P1B3 in Human Cancer Cell lines

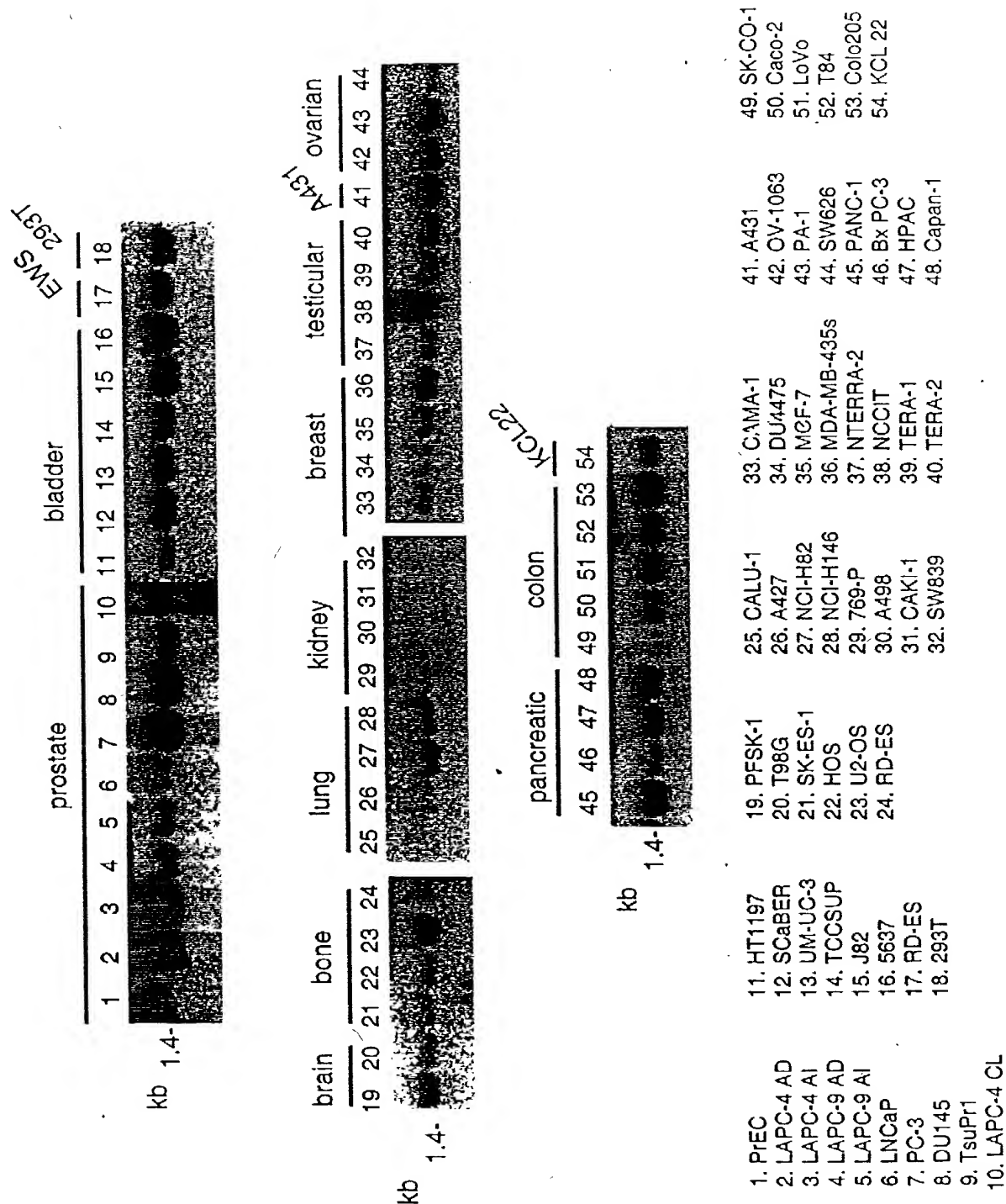
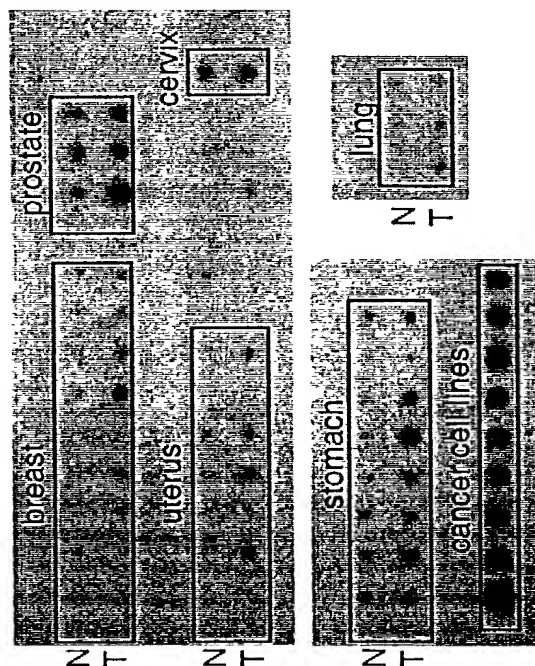


Figure 13 Expression of 85P1B3 in Patient Cancer Specimens and Cancer Cell Lines



T = tumor RNA

N = normal adjacent tissue RNA

Cancer cell lines are:
(from left to right)

HeLa (cervical carcinoma)

Daudi (Burkitt's lymphoma)

K562 (CML)

HL-60 (PML)

G361 (melanoma)

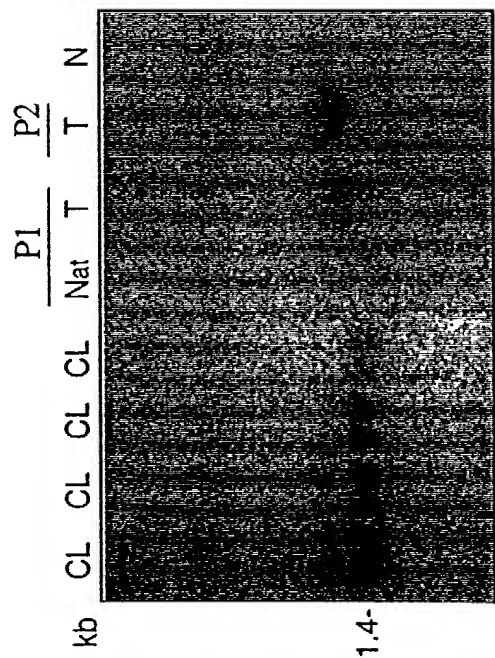
A549 (lung carcinoma)

MOLT-4 (lymphoblastic leuk.)

SW480 (colorectal carcinoma)

Raji (Burkitt's lymphoma)

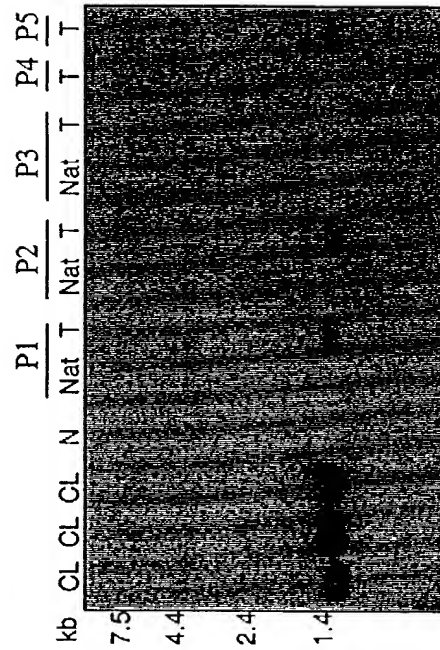
Figure 14 Expression of 85P1B3 in Colon Cancer Patient Specimens



Cell Lines (CL) - Colo 205, LoVo, T84, Caco-2
 N = Normal Colon
 Nat = Normal Adjacent tissue
 T = Tumor

P1 - Stage III, T2N1MX
 P2 - Stage III, T3N1MX

Figure 15 Expression of 85P1B3 in Bladder Cancer Patient Specimens

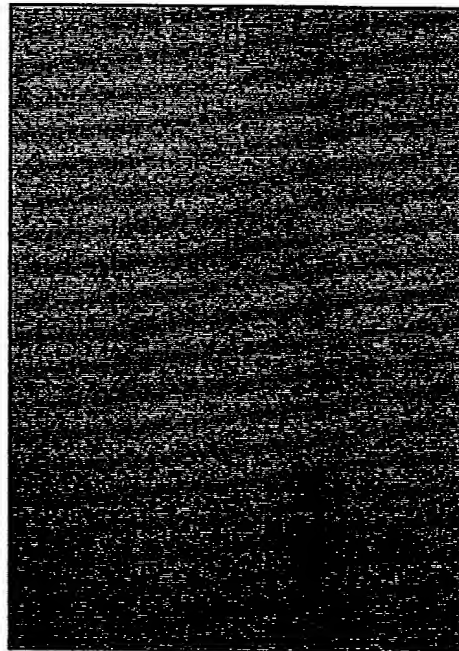


P1 - Transitional, grade 2
P2 - Transitional, grade 3/2
P3 - Transitional
P4 - Polypoid cystitis
P5 - Papillary, grade 3

CL = cell lines listed in order: UM-UC-3, J82, SCABER
P = Patient
N = Normal Bladder
Nat = Normal adjacent tumor
T = Tumor

Figure 16 Expression of 85P1B3 in Lung Cancer Patient Specimens

kb	CL	CL	CL	P1		P2		P3	
				NAT	T	NAT	T	NAT	T
9.5-									
7.5-									
4.4-									
2.4-									
1.4-									



P1 - Squamous, stage IB
P2 - Squamous, stage IIB
P3 - Squamous, stage IIIA

CL = cell lines (listed in order):
A427, NCI-H82, NCI-H146
P = Patient
N = Normal Lung
NAT = Normal adjacent tissue
T = Tumor

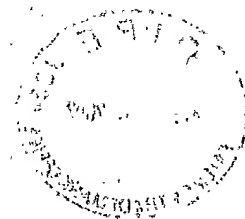
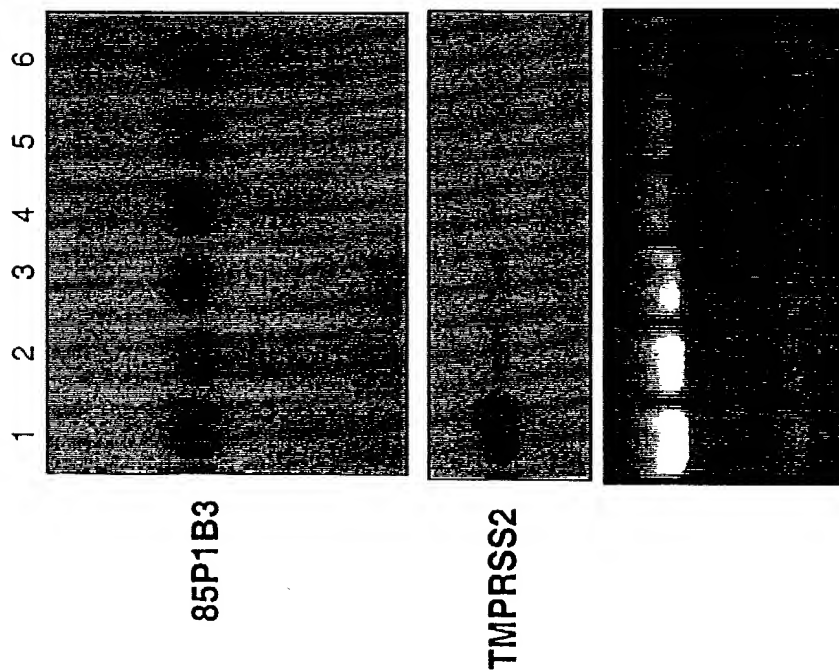


Figure 17 Expression of 85P1B3 in Prostate Cancer Xenografts Following
Castration



- LAPC-9AD Day 0 – Mouse #1
- LAPC-9AD Day 7 – Mouse #2
- LAPC-9AD Day 7 – Mouse #3
- LAPC-9AD Day 15 – Mouse #4
- LAPC-9AD Day 15 – Mouse #5
- LAPC-9AD Day 21 – Mouse #6

Figure 18 Expression of 85P1B3 in PC3 Cells Following Retroviral-Mediated Gene Delivery

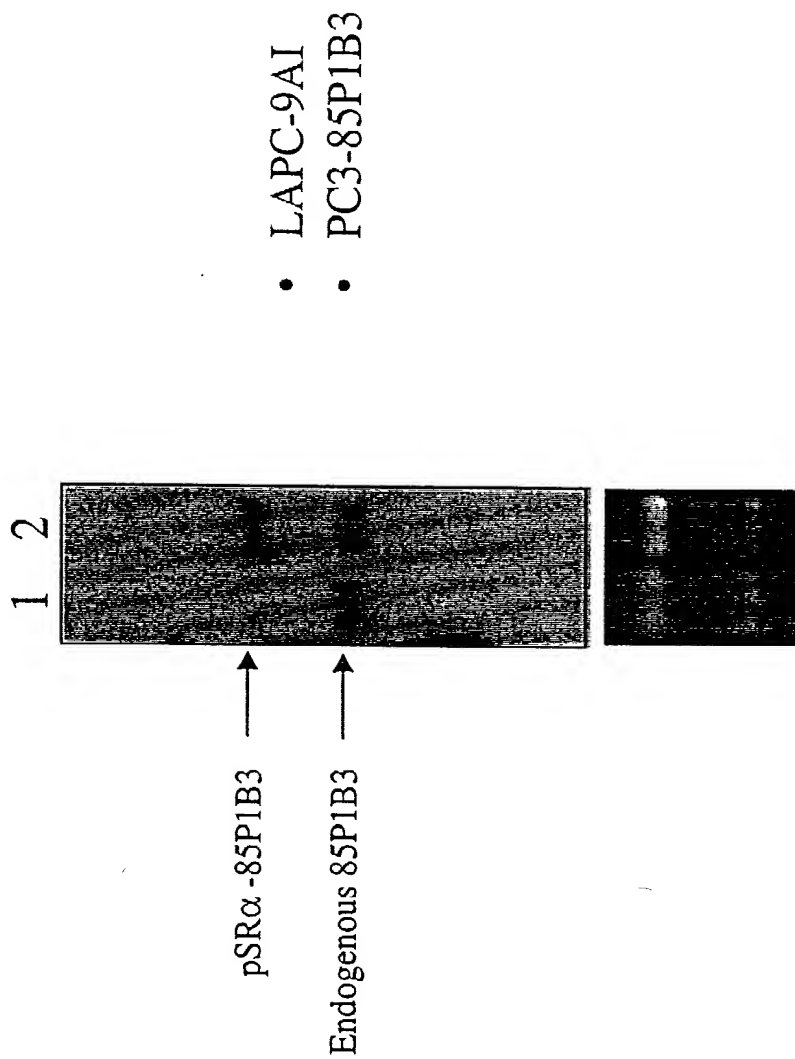
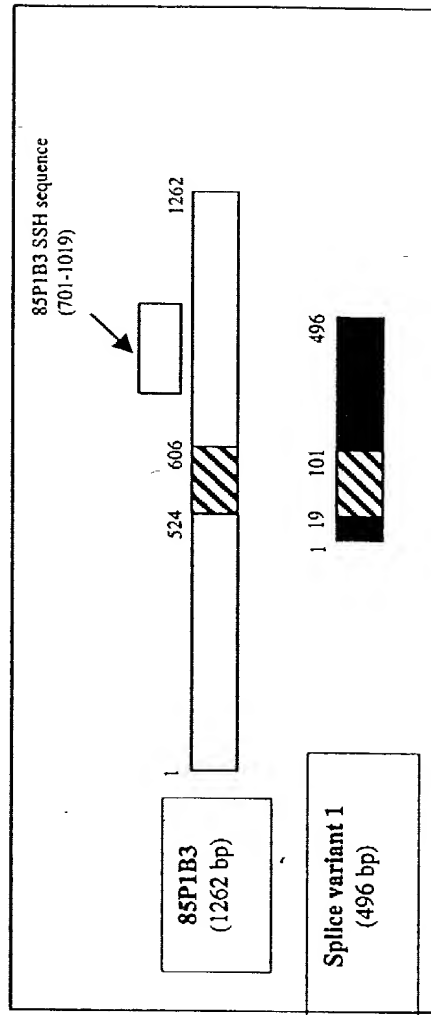
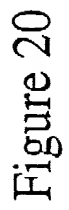




Figure 19.



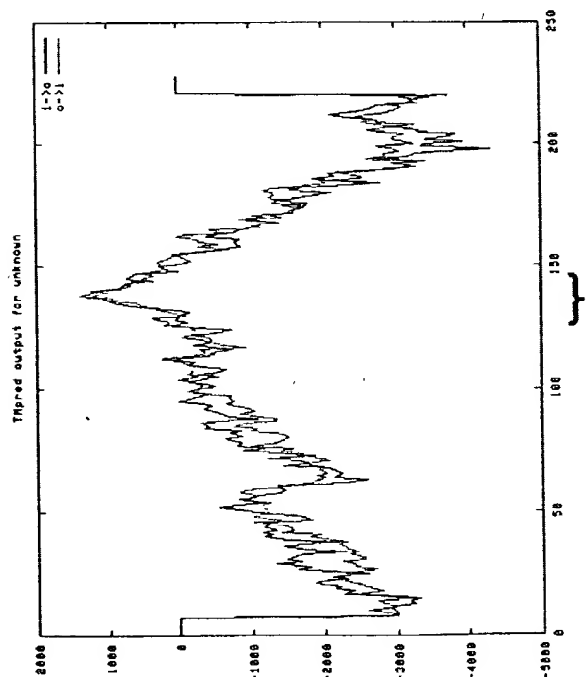


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10      |      20      |      30      |      40      |      50      |      60      |      70
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QPERCAVFQCAQCHAVLADSVHLAWDLRSILGAVFVSRTNNVLEAPFLVIGESLKGSTYNLLFCGSC
chhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhh
GIPVGFHLYSTHAALALRGHFCLSDDKMVCYLLKTKAIVNASEMDIQNVPLSEKIAELKEKIVLTHNRL
cccccccccccccccccccccccccccccccccccccccccccccccccccccccccccccccc
KSLMKILSEVTPDQSKPEN (SEQ ID NO:729)
hhhhhhhecccccccccccc

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Fig. 21B



transmembrane from amino acids 129-149

No transmembrane domains, soluble protein